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APPL-NO: JP53114091
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INT-CL (IPC): C09K 17/00

ABSTRACT:

PURPOSE: To obtain the title product useful for fertilization of soil, by adsorbing a culture of microorganisms to decompose and decay organic matter in soil and a specific substance necessary for them on vermiculite powder and calcium carbonate rock powder.

CONSTITUTION: (A) A culture of microorganisms, e.g. thermophlic fibrinolytic bacteria, actinomycetes (ray fungi), molds and yeasts, photosynthetic bactera, or heterotrophic bactera, useful for decomposition and decay of organic matter in soil as seed bacteria, in a medium and (B) a specific organic nitrogen source, vitamins, minor nutrients, and growth factors are adsorbed on (C) a mixture of vermiculite and calcium carbonate rock powder to give a soil activator.

EFFECT: High porosity, water and base retention improve the acid soil.

⑲日本国特許庁（JP）　⑪特許出願公開

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㊸公開　昭和55年(1980) 3月22日

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㊹土壌活性剤及びその製造方法

㉑特　願　昭53—114091
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(1)　(2)

明　細　書

1.発明の名称　土壌活性剤及びその製造方法

2.特許請求の範囲

(1).好熱性繊維分解菌、放線菌、糸状菌、酵母菌、光合成細菌、従属栄養細菌等の土壌中の有機性物質の分解、腐植化に役立つ微生物を種菌として、培地と共に、これ等の微生物の要求する特殊な有機性窒素源、ビタミン類、微量栄養素、微量生育因子等が、パーミキュライト粉と炭酸石灰岩粉との混和物に吸着されて成ることを特徴とする土壌活性剤。

(2).好熱性繊維分解菌、放線菌、糸状菌、酵母菌、光合成細菌、従属栄養細菌等の土壌中の有機性物質の分解、腐植化に役立つ微生物を種菌として、Viljoen、Prod、Peterson(1926)の培地または天然培地に、ペプトン、炭酸カルシウム過剰、リン酸水素アンモニウムナトリウム、リン酸二水素カリウム、硫酸マグネシウム、塩化カルシウム、塩化第二鉄痕跡、繊維素、井水または水道水或は他の清浄水等の水を使用し、60±5℃の嫌気的或は通性嫌気的条件下で、48～60時間培養し、これに、パーミキュライトと炭酸石灰岩粉とを加えてよく撹拌混合して吸着させることを特徴とする土壌活性剤の製造方法。

(3).前項発明における好熱性繊維分解菌、放線菌、糸状菌、酵母菌、光合成細菌、従属栄養細菌等の土壌中の有機性物質の分解、腐植化に役立つ微生物を種菌として、Viljoen、Prod、Peterson(1926)の培地または天然培地に、ペプトン、炭酸カルシウム過剰、リン酸水素アンモニウムナトリウム、リン酸二水素カリウム、硫酸マグネシウム、塩化カルシウム、塩化第二鉄痕跡、繊維素、井水または水道水或は他の清浄水等の水を使用し、60±5℃の嫌気的或は通性嫌気的条件下で、48～60時間培養し、これに、

パーミキュライトと炭酸石灰岩粉とを加えてよく攪拌混合して吸着させ、これを、粒状等の適宜形態に賦型することを　徴とする土壌活性剤の製造方法。

3 発明の詳細な説明

この発明は、土壌活性剤及びその製造方法の改良に係り、(1)．好熱性繊維分解菌、放線菌、糸状菌、酵母菌、光合成細菌、従属栄養細菌等の土壌中の有機性物質の分解、腐植化に役立つ微生物を種菌として、培地と共に、これ等の微生物の要求する特殊な有機性窒素源、ビタミン類、微量栄養素、微量生育因子等が、パーミキュライト粉と炭酸石灰岩粉との混和物に吸着されて成り、または、(2)．好熱性繊維分解菌、放線菌、糸状菌、酵母菌、光合成細菌、従属栄養細菌等の土壌中の有機性物質の分解、腐植化に役立つ微生物を種菌として、Viljoen, Prod, Peterson (1926) の培地または天然培地に、ペプトン、炭酸カルシウム過剰、リン酸水素アンモニウムナトリウム、リン酸二水素カリウム、硫酸マグネシウム、塩化カルシウム、塩化第二鉄溶液、微量素、井水または水道水或は他の清浄水等の水を使用し、60 ± 1℃の嫌気的或は通性嫌気的条件下で、48～60時間培養し、これに、パーミキュライトと炭酸石灰岩粉とを加えてよく攪拌混合して吸着し、或は、(3)．これを賦型するものであつて、好熱性繊維分解菌、放線菌、酵母菌、光合成細菌、従属栄養細菌等の土壌有効菌を人工培養し、これを種菌として散布、増殖し、有機性物質の分解腐植化を確実に、且つ促進して土壌の肥沃化をはかる土壌活性剤を得ようとすることを目的とするものである。

改めて指摘するまでもなく、わが国は、国土が狭く、資源も乏しい中にあつて、土は最も重要な資源であり、また、食糧経営の基盤でもある。しかし、

わが国の土壌は、多雨のために酸性化、塩基の流亡など自然条件の厳しさに加え、化学肥料と農薬偏重による多肥多収穫農法、更に、日夜集約化の方向を辿りつつある中で、労働力の不足等からの省力栽培化ということもあつて、土壌の劣悪化が著しく、気象災害等に対する抵抗力も弱まつて、地力の低下が強く懸念されている。この現実に対する強い反省が所轄官庁を始めとする関係諸機関の「土つくり運動」のより強力な推進である。

土づくりとは、植物が生育するための土地環境、または土壌の条件を満たし、その機能を最高の形にするための総合的努力であつて、結局、土づくりの目標は、良質の有機性物質の施用と深耕によつて、土壌微生物の働きを促がして、真正腐植を理学的にまた化学的に、そして生物的に安定した物質として土壌中に集積していくことである。

地力の根源は、土壌中の腐植である。腐植は有機窒素に富み、植物の養分である陽イオンの吸着保持、キレート作用、土壌の団粒化、微生物活性を促すなど、土つくりに欠くことのできない農業上きわめて重要な物質で、土壌の物理性、化学性は、土壌微生物活性に深く係つている。土壌には、屢々擬人法的表現がなされ、「土が生きている」とか、「土が疲れている」、「土が死んでいる」等といわれる。

土壌には、物質の形態を変化させる能力がある。この能力は、生物によつて引き起される化学変化なので、生化学的変化と云われ、この生化学的変化能力を土壌活性と呼んでいる。即ち、土壌活性は、微生物に由来することがはなはだ大きい。

従つて、本発明の目的は、好熱性繊維素分解菌、放線菌、酵母、光合成細菌、従属栄養細菌等の土壌有効菌を人工培養し、これを種菌として散布、増殖

し、有機性物質の分解腐植化をより確実に、且つより促進して土壌の豊かな肥沃化をはかろうとするものである。

次に、この発明の構成は、(I)．好熱性繊維素分解菌や紅色無硫黄細菌等の通性嫌気性または、嫌気性菌の培養、(II)．糸状菌、放線菌、酵母及びこの発明で使用する従属栄養細菌のような好気性菌の培養、(III)．特殊有機性窒素源のビタミン類及び微量生育因子の添加、(IV)．バーミキュライトと炭酸石灰岩粉との混合による賦型剤の製造と、前記微生物の培養との混和によるこの発明土壌活性剤の製造と云う4段階の要素的工程からできている。

中でも、この発明の特に強調したい特徴は、賦型剤としてバーミキュライトと炭酸石灰岩粉との混合物を使用したことである。

バーミキュライトは、次のような優れた性質を持つている。

(イ)．バーミキュライト

雲母別した蛭石（Vermiculite）を乾燥後、1000℃前後で焼成したものを、普通バーミキュライトと呼んでいる。

バーミキュライトの分析表

SiO_2 珪　酸	43.07 %
TiO_2 チタン	1.84
Al_2O_3 アルミナ	15.23
Fe_2O_3 酸化第二鉄	13.17
FeO　酸化第一鉄	1.08
MgO　苦土	7.16
CaO　石灰	2.01
K_2O　加里	3.32
$+H_2O$ 100℃で揮散しない結晶水	5.10
$-H_2O$ 100℃で揮散する水分	2.23
その他	3.07

の上記成分表は、その一例であつて、バーミキュライト自体のカリウムの含有量が多い。

(ロ)．気孔率が高く、水分吸収や保水力に優れ、排水や空気の流通がよく土壌団粒構造がよく発達するので、高度化した微生物の棲みかが豊富にできる。

(ハ)．著しく強力な塩基の置換性を持つているので、肥料持ちがよく、過剰肥料のコントロールに勝れた能力を持つている。

例えば、加里過剰による苦土欠乏症の防止に特異的な効果を示す。

(ニ)．栽培植物の発根が旺盛で、毛根がかつちりと、バーミキュライトに入り込むので、植え痛みが少ない。

好熱性繊維素分解菌等の通性嫌気性または、嫌気性菌の培養

(イ)．好熱性繊維素分解菌の培養

繊維素分解菌と称される中には、細菌、放線菌及び糸状菌等の種々の種類が含まれる。しかし、繊維素分解力の旺盛な点、幅広い繁殖条件などの点から植物性有機性物質の分解腐植化に Crostridium Thermocellum、Bacillus Thermocellulolyticus、Bacillus Thermofibrinolus、Bacillus Cellulosae dissolvens 等の好熱性細菌が重要な役割を果す。

好熱性繊維素分解菌の培養は、Viljoen、Fred、Peterson(1926) の培地：ペプトン 5 g、炭酸カルシウム過剰、リン酸水素アンモニウムナトリウム 2 g、リン酸二水素カリウム 1 g、硫酸マグネシウム 0.3 g、塩化カルシウム 1 g、塩化第二鉄痕跡、繊維素 15 g、井水または水道水 1000 cc を使用する。この培地組成の一部を天然物に置きかえてもよい。

60±5℃ 嫌気的或は、通性嫌気的条件下で 48 ～ 60 時間培養する。

(ロ)．紅色無硫黄細菌の培養

光合成細菌は、紅色硫黄細菌、緑色硫黄細菌及び紅色無硫黄細菌の三種類に大別される。この発明で、主として使用する細菌は、紅色無硫黄細菌で、本菌の持つ優れた性質、即ち有機性物質の分解によつて生ずる低分子の有機酸、アミノ酸、アルコール類等を好んで資化し、硫化水素を分解し、空気中の窒素を固定する能力等を積極的に活用する。

紅色無硫黄細菌の培養は、Hutner(1946)の培地、K_2HPO_4 0.05 (%)、KH_2PO_4 0.05 (%)、$(NH_4)_2HPO_4$ 0.05 (%)、$MgSO_4$ 0.02 (%)、乳酸 0.3 (%)、酢酸 0.1 (%)、クエン酸 0.1 (%)、Fe 200 (γ%)、Ca 500 (%)、B 5 (%)、Cu 1 (%)、Mn 100 (%)、Zn 200 (%)、Ca 1 (%)、Co 1 (%)、Mo 5 (%)

以上の成分を蒸留水に溶解し、更に、その1.000 ccにビオチン13.7 μg、酵母自己消化物600 mgを添加し、PHを6.8～8.3に調整したものを基本培地として使用する。そのときの状況に応じて、天然物に一部代 する。25±7℃、好気的または嫌気的(通性嫌気性)、明(光)または、 (光)の条件下で48～72時間培養する。

(ハ)・土記細菌の量産

一部天然物に代替するとともあるが、それぞれの単離または集殖用培地を使用する。好熱性枯草菌分解菌には、単段連続発酵方式により、また、紅色無硫黄細菌は、多段循環型連続発酵方式によつて、300～1000 l/日、嫌気的または通性嫌気的に多量培養する。

放線菌等の好気性菌の培養

(イ)・放線菌の培養

土壌中の働きについて一般的に云うことは、難しいが、各種の有機性物質、特に、難分解性のセルロース、リグニン等を分解し、土壌肥沃の下になる腐植の生成に他の微生物と共に重要な働きをしており、また、生物質の生産を通じてミクロフロラ・コントロールの面で重要な意義を持つものと見られる。

この発明で使用した放線菌は、主に、Actinomyces melanosporus型である。本菌の培養は、Krainsky (1914)の人工培地、塩化アンモニウム 0.05 g、リン酸水素二カリウム 0.05 g、寒天末 2.0 g、井水または水道水100 ccを用い、27±3℃、1～2週間保温。

(ロ)・糸状菌及び酵母菌の培養

便宜上、または実用上糸状菌と酵母菌とに大別されているが、系統分類学上、共に真正菌(Eumycetes)に属する。

糸状菌は、植物遺体などの有機性物質の分解に預かり、土壌の肥沃化に関係する。主として分解の初期段階に活動していると考えられる。

次に、酵母菌の土壌中における働きについては、不明な点が多い。しかし、土壌中には相当数の酵母菌が存在し、且つ、その保有する微量生長因子をめぐつて、他の微生物との共棒や、土壌活性など、将来の研究に期待されることが大きい。

糸状菌及び酵母菌の培養に、Czapek Dox(1910)の培地、硝酸ナトリウム 3 g、リン酸水素二カリウム 1 g、塩化カリウム 0.5 g、硫酸マグネシウム($MgSO_4 \cdot 7H_2O$) 0.5 g、硫酸第一鉄($FeSO_4 \cdot 7H_2O$) 0.01 g、蔗糖30 g(適宜)、蒸留水1000 cc、固型培地には寒天15 gを添加したものを使用する。

この発明では、糸状菌としてムコール属菌、アスペルス属菌、ペニシリウム属菌、トリコデルマ属菌等を、また酵母菌としては、ハンゼヌラ属菌、トルラ属菌、ピヒア属菌、エンドミセス属菌、サツカロミセス属菌等を土壌あるいは堆肥中より単離する。

(ハ)・従属栄養細菌(腐敗菌)の培養

類の分解も同様であるが、タンパク質を分解してアンモニヤを化成する細菌の特定のものは稀であつて、殆んど細菌一般の通性となつている。この発明では、好気性の枯草菌群細菌を利用する。

枯草菌群細菌の培養は、Wakeman(1922)の培地、ブドウ糖1g、リン酸水素二カリウム0.3g、硫酸マグネシウム($MgSO_4 \cdot 7H_2O$)0.2g、硫酸第二鉄($Fe_2(SO_4)_3 \cdot 9H_2O$)痕跡、卵白(粉末)0.25g、蒸留水1000cc、PH 7.2を使用して、本菌群を好気的に集殖する。

(ニ)．上記好気性菌の量産

単離または集殖培養した上記好気性菌を粗成細菌等を、10～20倍に稀釈した培地に接種し、800～1000 l/日、回分式(Batchwise)装置によつて、これに、滅菌空気を導入し、好気的条件下で多量培養する。

粗成細菌の成分の一例は、下記の通りであるが、必要があれば、窒素源またはリンの一部を添加する。

各種好気性菌の培地としては、比較的優秀であり、且つ安価で、経済的に菌を殖させることができる。

粗成細菌の成分

粗タンパク質	10.0 %
可溶性無窒素物	62.1 %
粗灰分	
カリウム	3.67 %
カルシウム	0.74 %
マグネシウム	0.33 %
ナトリウム	0.16 %
塩素・硫黄	微量
リン	0.08 %
ビタミン類	
ビタミンB_1	0.4 mg %
コーリン	560.0 mg %
パントテン酸	18.0 mg %
ナイアシン	20.0 mg %

リボフラビン・ピリドキシンが割合多く、ビタミンC・E等が若干

水分	26.0 %
可消化養分総量	54.0 %

特殊有機性窒素源、ビタミン類及び微量成育因子の添加

水田でも、畑地でも同様であるが、良質の耕作土中に$\times 10^7 \sim \times 10^8$と云う驚くべき数の細菌が存在する。その中で、糖と無機塩類だけで生育できるのは15%に満たない。大部分の細菌は、何んらかの形でアミノ酸、ビタミン類、VGF(未知の生育因子)を要求する。

好熱性嫌繊維素分解菌も、紅色無硫黄細菌も、またその例外ではない。若し、これらが欠除した場合、好熱性嫌繊維素分解菌の連続培養が不可能になり、また、紅色無硫黄細菌では増殖が停止して異常発酵を起す。

そこで、前者の微量生育因子をVGF－α、後者ではVGF－β(別名グロスター)とする。これらは、この発明者達が新規に発見したものであつて、VGF－αは、40 ppm以上、VGF－βは0.5 ppm以上を、それぞれの培養に使用する。

また、以上のような理由から、一般の土壌有効菌のために、下記のような微量栄養素をこの発明の土壌活性剤中に添加してある。

ビタミンB_1(チアミン)	1.00 ppm	以上
ビタミンB_2(リボフラビン)	5.00 〃	〃
ニコチン酸	800 〃	〃
ビタミンB_6(ピリドキシン)	0.40 〃	〃
パントテン酸	400 〃	〃
葉酸	0.20 〃	〃
コリン	10.0 〃	〃
ビオチン	0.20 〃	〃
ビタミンB_{12}(コバラミン)	0.03 〃	〃
パラアミノ安息香酸	5.00 〃	〃
コーンステンプリカ(CSL)	0.01 %	〃
脱脂大豆塩酸加水分解物	0.03 〃	〃

賦型剤と土壌活性剤の製造

先に、パーミキュライトの　性について、それ自体にカリウムの含量の多いこと、気孔率が高く、水分吸収や保水力に優れ、排水や空気の流通がよく、特に、強力な塩基の置換性を持つていること等を上げたが、炭酸石灰岩粉についても、また、カルシウムイオンやマグネシウムイオンは好熱性纖維素分解菌を始め、土壌有効菌の栄養源となるばかりでなく、土壌水素イオン濃度の調整や土壌団粒構造の造成、その他の良好な環境条件を作るのに役立つものである。

従つて、両者の特性と、耕地の利用法、土壌の性質、或は栽培植物の種類等に応じて、炭酸石灰岩粉に対して10%から50%まで、両者の配合割合と、更に散布器機の種類等によつて両者の粒度を定め、最後に有害菌の汚染、保存、工程管理、経済性、種菌劣化の防止　まで考慮し、総合的な判断の下に粉末状、ペリット状、パール状　土壌活性剤の形態を決定する。

そこで、この発明は、前記の通り、土壌中の有機性物質の完熟腐植化に役立つ菌、即ち好熱性纖維素分解菌及び紅色無硫黄細菌等の嫌気的または通性嫌気的培養に、それぞれ適合する天然高分子、凝集剤を加えて得られる濃厚菌体液、放線菌、糸状菌、酵母菌、従属栄養細菌等の好気的粗成糖蜜培養を培地と共に、更に有機性窒素源、ビタミン類、微量生育因子等を、パーミキュライトと炭酸石灰岩粉とを主材とした賦型剤に加えて、よく撹拌混合し、決定された形態の製品とする。

原材料配合の一例は、下記の通りである。

原材料の配合割合

（炭酸石灰岩粉1,000 gに対して）

好熱性纖維素分解菌の濃厚菌体液	0.2g
紅色無硫黄細菌の濃厚菌体液	0.3 g
放線菌、糸状菌、酵母菌、従属栄養細菌の粗成糖蜜	
培養液	5.0 g
VGF－α	55.0 mg
VGF－β（別名グロスター）	15.0 mg
ビタミンB_1	1.2 mg
〃　B_2	5.5 mg
ニコチン酸	830.0 mg
ビタミンB_6	0.5 mg
パントテン酸	820.0 mg
葉　酸	0.3 mg
コリン	12.0 mg
ビオチン	0.2 mg
ビタミンB_{12}	0.1 mg
パラアミノ安息香酸	7.0 mg
コーンステップリカ（CSL）	0.3 g
脱脂大豆塩酸加水分解液	0.7 g
パーミキュライト	200 g
炭酸石灰岩粉	1,000 g

このようにして、この発明の優れた効果として、次のような利点を挙げることができる。

(イ)．好熱性纖維素分解菌、放線菌、糸状菌、紅色無硫黄細菌、酵母菌、従属栄養細菌のような土壌有効菌を培養し、これらを人為的に土壌中に添加して、菌の密度を高めることは、現在、日本農業の「土つくり」に対して、著しく有効な一つの方法である。

(ロ)．このような人工接種法が成功するか否かは、菌が定着し、活動する条件が造れるかどうかにかかるが、同時に多量に散布される賦型剤のパーミキュライト及び炭酸石灰岩粉は、排水、通気、水分吸収、保水、団粒構造の造成、水素イオン濃度の調整等高度化した微生物の棲みかを豊富に造ると共に、栽培植物の土壌環境条件を改善するのに役立つ。

(ハ)．土壌微生物の必要とする各種微量栄養素の添加及び紅色無硫黄細菌と酵母菌の増殖は、土壌微生

物社会のサクセションがうまく行われて、土壌有機性物質の真正腐植化が確実且つ、迅速に行われる。

(ヨ)．また、種菌を固型状とし、粉末、ペリット、パール状とその形態を選ぶことによつて、その保存性、散布等を容易、確実なものにする。なお、この発明の土壌活性剤は、低温度、冷暗所等の比較的保存条件のよい所では、数年間、種菌の有効性を保持する。

この発明による土壌活性剤を施用した実施例のいくつかでは、そのすばらしい効果を更によく実証するものである。

実施例1

堆肥は、「土づくり」のための最高の総合的効果の高い資材である。この発明の土壌活性剤は、堆肥の熟成にもすばらしい効果を示す。

イネワラ1,000 Kgに対して60 Kgの粉末状土壌活性剤（バーミキュライト：炭酸石灰岩粉＝20：100）と水分を加えて、約10日間仮積する。次に、窒素1.2 Kgに相当する硫安または、尿素を散布し、適度に散水しながら軽く踏み付けながら本積とする。途中、1回切り返しを行う。よく発酵し40日で完了する。

よく腐熟し、イネワラは容易にちぎれる程度となり、炭素率は17.3を示す。

そして、この発明の土壌活性剤の代りに、バーミキュライトと炭酸石灰岩粉（比率＝20：100）の混合物50 Kgを加えたものと無添加のものとを対照とし、その他は、土壌活性剤施用のものと全く同様にして平行実施した結果は、前者は半熟程度、後者は堆肥は認められなかつた。なお、対照前者の炭素率は31.8後者は37.2であつた。

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SOIL ACTIVATOR AND ITS METHOD OF MANUFACTURE
[DOJO KASSEIZAI OYOBI SONO SEIZO HOHO]

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Specifications /167*

1. Title of the Invention

Soil Activator and Its Method of Manufacture

2. Claim(s)

(1) A soil activator characterized by being comprised by making an inoculum from microorganisms, such as thermophilic fibrinolytic bacteria, Actinomycetes, filamentous bacteria, yeast fungi, photosynthetic bacteria, or heterotrophic bacteria, which are useful for decomposing and humifying of organic substances in the soil, culturing them on a culture medium, and specific organic nitrogen sources, vitamins, minor nutrients, minor growth factors needed by these microorganisms being adsorbed on a mixture of vermiculite and calcium carbonate rock powder.

(2) A method for manufacturing a soil activator characterized by making an inoculum from microorganisms, such as thermophilic fibrinolytic bacteria, Actinomycetes, filamentous bacteria, yeast fungi, photosynthetic bacteria, or heterotrophic bacteria, which are useful for decomposing and humifying organic substances in soil using peptone, excess calcium carbonate, ammonium and sodium hydrogen phosphate, potassium dihydrogen phosphate, magnesium sulfate, calcium chloride, ferric chloride, fibrin, and water, such as well water, tap water or other clean water, in a Viljoen, Fred, Peterson (1926) culture medium, or natural culture medium, then culturing this for 48 to 60 hours under anaerobic or facultative anaerobic conditions at 60±5°C, adding vermiculite and calcium carbonate rock powder to this, and stirring this well to mix and adsorb it thereon.

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* Numbers in the margin indicate pagination in the foreign text.

(3) A method for manufacturing a soil activator characterized by making an inoculum from microorganisms, such as thermophilic fibrinolytic bacteria, Actinomycetes, filamentous bacteria, yeast fungi, photosynthetic bacteria, or heterotrophic bacteria, which are useful for decomposing and humifying organic substances in soil using peptone, excess calcium carbonate, ammonium and sodium hydrogen phosphate, potassium dihydrogen phosphate, magnesium sulfate, calcium chloride, ferric chloride, fibrin, and water, such as well water, tap water or other clean water, in a Viljoen, Fred, Peterson (1926) culture medium, or natural culture medium, then culturing this for 48 to 60 hours under anaerobic or facultative anaerobic conditions at 60±5°C, adding vermiculite and calcium carbonate rock /168 powder to this, stirring this well to mix and adsorb it thereon, and shaping this into a suitable form, such as a granular form.

3. Detailed Specifications

This invention pertains to improving a soil activator and its method of manufacture, by (1) making an inoculum from microorganisms, such as thermophilic fibrinolytic bacteria, Actinomycetes, filamentous bacteria, yeast fungi, photosynthetic bacteria, or heterotrophic bacteria, which are useful for decomposing and humifying of organic substances in the soil, culturing them on a culture medium, and specific organic nitrogen sources, vitamins, minor nutrients, minor growth factors needed by these microorganisms being adsorbed on a mixture of vermiculite and calcium carbonate rock powder, (2) by making an inoculum from microorganisms, such as thermophilic fibrinolytic bacteria, Actinomycetes, filamentous bacteria, yeast fungi, photosynthetic bacteria, or heterotrophic bacteria,

which are useful for decomposing and humifying organic substances in soil using peptone, excess calcium carbonate, ammonium and sodium hydrogen phosphate, potassium dihydrogen phosphate, magnesium sulfate, calcium chloride, ferric chloride, fibrin, and water, such as well water, tap water or other clean water, in a Viljoen, Fred, Peterson (1926) culture medium, or natural culture medium, then culturing this for 48 to 60 hours under anaerobic or facultative anaerobic conditions at 60±5°C, adding vermiculite and calcium carbonate rock powder to this, and stirring this well to mix and adsorb it thereon plus (3) shaping this into a suitable form, such as a granular form, and the object is to obtain a soil activator for artificially culturing bacteria which is effective on soil, such as, thermophilic fibrinolytic bacteria, Actinomycetes, yeast fungi, photosynthetic bacteria, or heterotrophic bacteria, and scattering and propagating this as an inoculum, to reliably promote the decomposition and humification of the organic substances and to plan fertilization of soil.

It is not necessary to point out yet again that Japan has few resources, and that the soil is our most important resource, and is the foundation of farming. However, besides the harsh natural conditions in Japan, such as acidification of soil due to downpours and runoff of bases, the soil in Japan is sometimes cultivation with insufficient labor due to a shortage of labor or the like while guiding the direction of high-fertilization and high-harvesting farming methods by chemical fertilizers and [illegible] performed intensely day and night, there is strong concern that the soil will become inferior, the soils resistance against weather and fire disasters

or the like also will weaken, and the soil's fertility will decrease. Intense contemplation with respect to this reality is a powerful driving force in a "movement for soil preparation" by the many concerned institutions like local jurisdictional government offices.

Soil preparation is a synthetic effort for satisfying the environment or conditions for soil for growing plants to maximize its function. A final goal of soil preparation is to stimulate the activity of microorganisms in the soil by the application and deep plowing of good-quality organic substances, and a genuine humus is accumulated in the soil as a physically-, chemically-, and biologically-stabilized substance.

The root of soil fertility is the humus therein. Humus is rich in organic nitrogen and is an extremely important agricultural substance because it promotes the adsorption and retention of cations, which are mineral ameliorants, a chelating action, the granulating of soil, and microbial activity without compromising the soil preparation; the physical and chemical properties are closely related to the activity of the microorganisms in the soil. Various personifications of soil have been implemented, such as "the soil is alive," "the soil is tired," "the soil is dead," etc.

There is an ability to change the form of the substances in the soil.

This ability is a chemical change is induced by organisms, referred to as a biochemical change. This biochemical-changing ability is called soil activity. That is, soil activity is most often derived from microorganisms.

Therefore, the object of the present invention is to synthetically culture bacteria which are effective in soil, such as thermophilic fibrinolytic bacteria, Actinomycetes, yeast fungi, photosynthetic bacteria, or heterotrophic bacteria and scatter and propagate them as an inoculum to accelerate the decomposition and humification of organic substances more reliably and plan richer fertilization of the soil. /169

Next, the constitution of this invention comprises four fundamental steps, i.e., (I) culturing facultative anaerobic or just anaerobic bacteria, such as thermophilic fibrinolytic bacteria or Rhodospirillaceae, (II) culturing anaerobic bacteria, such as filamentous bacteria, Actinomycetes, yeast fungi, or heterotrophic bacteria, (III) adding vitamins and minor growth factors as specific organic nitrogen sources, and (IV) manufacturing the soil activator of this invention by mixing the manufacture of an excipient, by mixing vermiculite and calcium carbonate rock powder, with the culturing of the aforesaid microorganism.

The specific feature of this invention that is emphasized is that a mixture of vermiculite and calcium carbonate rock powder is used as the excipient.

Vermiculite has the following superior properties.

(a) Vermiculite

Sieved vermiculite that is dried and subsequently baked around 1,000°C is referred to as regular vermiculite.

Analysis Table of Vermiculite

SiO	Silicic Acid	45.07%
TiO_2	Titanium	1.84
Al_2O_3	Alumina	15.25
Fe_2O_3	Ferric oxide	13.17
Feo	Ferrous oxide	1.08
MgO	Magnesia	7.16
CaO	Lime	2.01
K_2O	Potash	3.32
$+H_2O$	Nonvolatile crystal water at 100°C	5.80
$+H_2O$	Volatile moisture at 100°C	2.23
Other		3.07

The table with the above constituents is one example of vermiculite, and the potassium content of the vermiculite per se is high.

(b) Since the porosity is high, the moisture absorption and water holding capacity are excellent, and the drainage and air distribution is developed well, only sophisticated microorganisms are enriched.

(c) Since it has a remarkably powerful substitutability for bases, its fertilizer-holding ability is good, and it has a superior ability to control excess fertilizer.

For example, it exhibits a unique effect in preventing Wakatsuchi deficiency caused by excess potash.

(d) Rooting of cultured plants is vital, and if their hair roots are solidly established in it, they will penetrate into the vermiculite; hence, the plants are hurt little.

Facultative anaerobic, or just aerobic culturing of thermophilic fibrinolytic bacteria

(a) Culturing of thermophilic fibrinolytic bacteria

Fibrinolytic bacteria include various kinds of bacteria, such as Actinomycetes and filamentous bacteria. However, in terms of the vitality of the fibrinolytic ability, the wide-ranging breeding conditions, and

the like, thermophilic bacteria, such as Crostridium Thermocellum, Bacillus Thermocelluloytieus, Bacillus Thermofibrincolus, and Bacillus Celulosae dissolveus, play important roles in the decomposition and humification of vegetable organic substances.

For culturing thermophilic fibrinolytic bacteria, 5 g of peptone, excess calcium carbonate, 2 g of ammonium and sodium monohydrogen phosphate, 1 g of potassium dihydrogen phosphate, 0.3 g of magnesium sulfate, 1 g of calcium chloride, 15 g of fibrin, and 1,000 cc of well water or tap water are used for a Viljoen, Fred, Peterson (1926) culture medium. Part of this culture medium composition may be replaced with natural materials.

The bacteria are cultured for 48 to 60 hours under 60±5 °C facultative anaerobic conditions.

(b) Culturing Rhodospirillaceae

Photosynthetic bacteria are roughly classified into three types: /170 Chlorobiceae, Chloroflexaceae, and Rhodospirillaceae. The bacteria used mainly in this invention are Rhodospirillaceae. The low-molecular weight organic acids, amino acids, alcohols, and the like produced by the superior properties of these bacteria, that is, the decomposition of the organic substances, are assimilated well, hydrogen sulfide is decomposed, and the ability for fixing nitrogen from the air, and the like is put to practical use proactively.

For culturing Rhodospirillaceae, a Huimer (1946) culture medium, in which the following constituents were dissolved in distilled water, i.e., K_2HPO_4 0.05(%), KH_2PO_4 0.05(%), $(NH_4)_2HPO_4$ 0.08(%), $MgSO_4$ 0.02(%), lactic acid 0.3(%), acetic acid 0.1(%), citric acid 0.1(%), Fe 200(γ%), Ca 500(%),

B5(%), Cu 1(%), Mn 100(%), Zn 200(%), Ga 1(%), Co 1(%), Mo 5(%), then 13.7 kg of biotin and 600 mg of yeast fungi self-digestible materials were added to 1,000 cc of this solution, and the pH was adjusted to 6.8 to 8.5 is used as the basal medium. At that time, the constituents are partially substituted with natural substances, depending on the circumstance. The bacteria are cultured for 48 to 72 hours at 25±7°C, under aerobic or anaerobic (facultative anaerobic), and light or dark conditions.

(c) Mass production of above-mentioned bacteria

Although constituents can be partially substituted with natural substances, the respective isolation or proliferative culture medium is used. A large amount of thermophilic fibrinolytic bacteria are cultured anaerobically, or facultative anaerobically by a single step continuous fermentation system, and moreover, a large amount of Rhodospirillaceae are cultured by a multistep circulation-type continuous fermentation system at a rate of 300 to 1,000 L/day.

Culturing of aerobes, such as Actinomycetes

(a) Culturing of Actinomycetes

Although difficult generally speaking, Actinomycetes has an important function, along with the other microorganisms, for decomposing various organic substances, and in particular, cellulose, lignin, and the like which are difficult to decompose, and producing humus under fertility of soil. Moreover, it is seen that it is important in the sense of microflow control through the production of biomaterials.

Actinomycetes used in the invention is primarily Actinomycetes melanosporus. Culturing of this bacterium is performed by using a Krainsky

(1914) synthetic culture medium consisting of 0.05 g of ammonium chloride, 0.05 g of potassium dihydrogen phosphate, 2.0 g of fibrin, and 100 cc well water or tap water, and maintaining the temperature for 1 to 2 weeks at 27±3°C.

(b) Culturing of filamentous bacteria and yeast fungi

Although filamentous bacteria and yeast fungi are general classifications as a matter of convenience and practical use, both of these belong to the phylum Eumycetes in terms of a systematic taxonomy.

Filamentous bacteria are entrusted to the decomposition of organic substances, such as vegetable remains, which is related to the fertilization of soil. It is thought that they primarily act in the initial step of decomposition.

Next, the function of yeast fungi in soil is often unclear. However, a considerable number of yeast fungi exist in soil and, and their [illegible] with other microorganisms that compete with the minor growth factor they possess, activity in soil, and the like are highly anticipated with future research.

Culturing of filamentous bacteria and yeast fungi is performed on a Czapek Dox (1910) culture medium containing 2 g of sodium nitrate, 1 g of potassium dihydrogen phosphate, 0.5 g of potassium chloride, 0.5 g of magnesium sulfate ($MgSO_4 \cdot 7H_2O$), 0.01 g of ferrous sulfate ($Fe\ SO_4 \cdot 7H_2O$), 30 g of sucrose (suitable), and 1,000 cc of distilled water; 15 g of agar added as a solid medium.

In this invention, filamentous bacteria, such as Mucor fragilis, Aspersus [transliteration], Penicillium spp., and Trichoderma, are isolated

in soil or compost, and yeast fungi, such as Hansenula, Torula, Endomyces, and Saccharomyces, are isolated therein.

(c) Culturing heterotrophic bacteria (putrefying bacteria)

As with decomposition of sugars, specific bacteria that break /171 down proteins and transform ammonia are rare, and are generally facultative on most bacteria. In this invention, aerobic Bacillus subtilis group bacteria are utilized.

Culturing of Bacillus subtilis group bacteria is performed in a Waksman (1922) culture medium of 1 g of glucose, 0.5 g of potassium dihydrogen phosphate, 0.2 g of magnesium sulfate ($MgSO_4 \cdot 7H_2O$), trace ferrous sulfate ($Fe_2(SO_4)_3 \cdot 9H_2O$), 0.025 g of egg white (powder), and 1,000 cc distilled water, at a pH of 7.2, and this bacteria group is proliferated aerobically.

(d) Mass production of above-mentioned aerobic bacteria

A culture medium diluted 10- to 20-fold is inoculated with the above-mentioned aerobic bacteria subjected to an isolated or collected culturing with a crude syrup, sterilized air is introduced into this using an 800 to 1,000 L/day, batch device, and a large amount is cultured under aerobic conditions.

An example of crude syrup constituents are as follows, but part of the nitrogen source or phosphorus is added, as needed.

Relatively superior bacteria may be propagated inexpensively and economically in the culturing of various aerobic bacteria.

Constituents of crude syrup

Crude protein	10.0%
Soluble nitrogen-free material	62.1%
Crude ash	
Potassium	3.67%
Calcium	0.74%
Magnesium	0.35%
Sodium	0.16%
Chlorine/sulfur	Tiny amount
Phosphorus	0.08%
Vitamins	
Vitamin B_1	0.4 mg%
Choline	860.0 mg%
Pantothenic acid	18.9 mg%
Niacin	20.0 mg%
Riboflavin/pyridoxine ratio	Large
Vitamins C, E, etc.	Small
Moisture	26.0%
Total digestible ameliorants	54.0%

Addition of special organic nitrogen sources, vitamins, and minor growth factors

An amazing number of bacteria on the order of $\times 10^7$ to $\times 10^9$ are present in a good-quality plowed layer, as in a paddy field or farmland. Only 15% of these bacteria are able to grow in sugar and inorganic salts. The majority of the bacteria require some form of amino acid, vitamin,

and VGF (unidentified growth factor).

Both thermophilic fibrinolytic bacteria and Rhodospirillaceae are no exception to this. Supposing these bacteria are depleted, continuous culturing of thermophilic fibrinolytic bacteria becomes impossible and propagation of Rhodospirillaceae is suspended, so an abnormal fermentation occurs.

Therefore, the first minor growth factor is VGF-α and the latter is VGF-β (alias: Gloucester). These new growth factors were discovered by the inventors of this invention. 40 ppm or more of VGF-α and 0.5 ppm or more of VGF-β are respectively used for culturing.

Moreover, according to the reasons described above, for general bacteria which are effective in soil, the following minor nutrients are added to the soil activator of this invention.

Vitamin B_1 (thiamine)	1.00 ppm or more
Vitamin B_2 (riboflavin)	5.00 〃 〃
Nicotinic acid	800 〃 〃
Vitamin B_6 (pyridoxine)	0.40 〃 〃
Pantothenic acid	400 〃 〃
Folic acid	0.20 〃 〃
Choline	10.0 〃 〃
Biotin	0.20 〃 〃
Vitamin B_{12} (cobalamin)	0.05 〃 〃
Paraamino benzoic acid	5.00 〃 〃
Corn steep liquor (CSL)	0.01% 〃 〃
Defatted soybean hydrochloric acid hydrolysate	0.03% 〃 〃

Manufacture of Excipient and Soil Activator /172

The physical properties of vermiculite include its large potassium content, high porosity, excellent moisture absorption and holding ability, good drainage and air circulation, and particularly powerful substitutability of bases, etc. But not only do the calcium carbonate rock powder, calcium ions and magnesium ions become nutrient sources for effective bacteria on soil, such as thermophilic fibrinolytic bacteria, they are useful for adjusting the hydrogen ion concentration in soil and creation of a granulated soil structure and for making conditions for a satisfactory soil environment.

Therefore, the particle sizes of the excipient and soil activator are determined according to their physical properties, their compounding ratios, which are from 10% to 50% of the calcium carbonate rock powder, depending on the method of using arable land, the properties of the soil, the type of cultivated plant, or the like, the type of spreader, etc. Lastly, the form of the soil activator, such as powdered, pellet-shaped, or pearl-shaped, is determined under an integrated determination by considering the contamination, preservation, process control, economics, deterioration and extermination of harmful bacteria and inoculum thereof.

Therefore, in this invention, as described above, in addition, to an activator composed mainly of the organic nitrogen source, vitamins, minor growth factors, and the like, as well as the vermiculite and calcium carbonate rock powder, upon stirring and mixing these well and using a culture medium, a product with a predetermined shape is obtained from an aerobic crude syrup culture of a concentrated microbial cell fluid

is obtained by adding a respectively conforming natural polymer and flocculant, Actinomycetes, filamentous bacteria, yeast fungi, and heterotrophic bacteria, for the anaerobic and facultative anaerobic culturing of bacteria useful for aging and humifying organic substances in the soil, i.e., thermophilic fibrinolytic bacteria, Rhodospirillaceae, etc.

An example of compounding raw materials is as follows.

Compounding Ratio of Raw Materials

(in 1,000 g of calcium carbonate rock powder)

Conc. thermophilic fibrinolytic bacteria microbial cell fluid	0.2 g
Conc. Rhodospirillaceae microbial cell fluid	0.5 g
Crude syrup of Actinomycetes, filamentous bacteria, yeast fungi and heterotrophic bacteria	5.0 g
Culture fluid	55.0 g
VGF-α	15.0 mg
VGF-β (alias: Gloucester)	1.2 mg
Vitamin B_1	1.2 mg
" " B_2	5.5 mg
Nicotinic acid	830.0 mg
Vitamin B_6	0.5 mg
Pantothenic acid	420.0 mg
Folic acid	0.3 mg
Choline	12.0 mg
Biotin	0.2 mg
Vitamin B_{12}	0.1 mg
Paraaminobenzoic acid	7.0 mg
Corn steep liquor (CSL)	0.3 mg
Defatted soybean hydrochloric acid hydrolysate	0.7 mg
Vermiculite	200 g
Carbon rock powder	1,000 g

The following merits may be cited as the superior advantages of this invention as such.

(a) Culturing bacteria which are effective on soil, such as thermophilic fibrinolytic bacteria, Actinomycetes, filamentous bacteria, Rhodospirillaceae, yeast fungi, or heterotrophic bacteria, and improving

the density of the bacteria by adding them to the soil artificially is currently one remarkably effective method for an agricultural soil preparation in Japan.

(b) Whether or not such an artificial inoculating method is successful and whether the conditions for fixing and activating the bacteria are established, the vermiculite and the calcium carbonate rock powder of the excipient scattered in large amounts simultaneously play a role for enriching the [illegible] of sophisticated microorganisms, such as the drainage, circulation, absorption of moisture, holding of water, creation of a granulated structure, and adjustment of the hydrogen ion concentration, and at the same time, for improving the conditions for the soil environment for cultivated plants.

(c) The addition of various minor nutrients required of microorganisms in the soil and the propagation of Rhodospirillaceae and yeast fungi /173 are performed with good succession in a soil microorganism system, and the genuine humification of organic substances in the soil is performed reliably and rapidly.

(d) Moreover, the preservation, scattering, and the like of a solid inoculum are easy and reliable by selecting the form thereof, such as powdered, pellet-shaped or pearl-shaped. Moreover, the validity of the inoculum of the soil activator of this invention is maintained for several years in places with relatively good preservation conditions, such as places with low humidity, cold and dark places, etc.

The superb advantages of the soil activator will be further demonstrated in a few practical examples in which it is applied according to this invention.

Practical Example 1

Compost is a raw material with the maximum integrated effects for "soil preparation." The soil activator of this invention also exhibits a superb advantage for aging compost.

60 kg of a powdered soil activator (vermiculite: calcium carbonate rock powder = 20:100) and moisture were added to 1,000 Kg of rice straw and temporarily heaped for about 10 days. Next, ammonium sulfate or urea equivalent to 1.2 kg of nitrogen was scattered thereon to make a main pile while sprinkling water on it and lightly trampling it properly. This is repeated once halfway through the procedure. The compost is fermented completely in 45 days.

The compost is aged well to the extent that the rice straw can be torn into pieces readily and the carbon rate is 17.3.

Then, as a result of preparing controls with and without adding 50 kg of a mixture of vermiculite and calcium carbonate rock powder (ratio=20:100) instead of the soil activator of this invention, and applying them concurrently in the same way as the method for applying a soil activator, the compost in the first control was semi-aged and no compost was verified in the latter control. Moreover, the carbon rate of the first control was 31.8, while that of the latter was 37.2.